

**Remarks / Arguments**

The claims were presently redrafted (see new claims 14-24). It is respectfully submitted that the amendment does not contain new matter, as indicated in the table below:

New claim	Basis for amendment
14	Claim 1 and Page 1, lines 27-36
15	Claim 2
16	Claim 3 and Page 1, lines 27-31
17	Claim 4
18	Claim 5 and Page 1, lines 33-36
19	Claim 6
20	Claim 7
21	Claim 1 and Page 1, lines 27-36
22	Claim 3
23	Claim 5
24	Claim 5 and Page 13, lines 10-13

**Claims 1-8 are rejected under 35USC102 as being anticipated by Nickel (US patent 5,034,227).**

The claims were redrafted, and now recite that the pieces of equipment are in *a configuration used in a potato starch factory*.

Nickel does not teach such a configuration. Moreover, Nickel does not disclose the combination of features recited in new claim 24.

In addition, it is clear to those skilled in the art, that the configuration used in a starch factory greatly differ according to the nature of the plant being processed.

In this respect, the Applicant herewith provides the following documents (see Appendixes 1-3):

- Appendix 1: Vasanthan et al., shows that the native starch granules greatly differ in size and morphology from one plant to another.
- Appendix 2: from the USIPA website, teaches that "*the manufacturing processes are specific for each plant, and the industrial tools are*

*dedicated to one raw material (wheat, corn, or potato in Europe),  
without it being possible to substitute one with each other”*

- Appendix 3: from STARCH, Whistler et al, describes the processing of corn (page 30), wheat (page 58), and potato (page 93).

The Applicant thus respectfully submits that the subject-matter of the pending claims is neither taught, nor suggested by the prior art.

In view of the above, it is respectfully submitted that the application is now in proper form for allowance.

Respectfully submitted,  
Young & Thompson



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Robert J. Patch, Reg. No. 17,355  
745 S. 23<sup>rd</sup> St.  
Arlington, VA 22202  
703-521-2297

RJP/fb

## **Translation of Appendix 2**

### Extraction processes

The craft in the starch and potato starch industry consists in separating the plant constituents: starch, proteins, cellulose envelopes, soluble fractions; and for corn, germs, from which oil is extracted. This first phase involves a series of simple steps for physically separating the constituents: grinding, sieving, centrifuging, etc... However, the manufacturing processes are specific for each plant, and the industrial tools are dedicated to one raw material (wheat, corn, or potato in Europe), without it being possible to substitute one with each other. In a second phase, the starch, thus extracted in its purest form, is either used as such after drying ("native starch"), or sent to numerous "workshops", wherein it is processed so that either its performances are improved ("modified starches") or one obtains glucose syrups. The latter in turn lead to multiple finished products... but that is another story!

In the end, starch and derived products are delivered to the client industries in the form of powder (like flour) or in the form of syrups.

In the French language, as opposed to the English language, wherein only one term is used (starch), starch is called "féculé" when it is extracted from tubers or roots (potatoes, manioc, or sweet potatoes).

\* \* \*

# The Reactivity of Native and Autoclaved Starches from Different Origins Towards Acetylation and Cationization

T. Vasanthan, F.W. Sosulski, Saskatoon (Canada), and R. Hoover, St. John's, Newfoundland (Canada)

Native potato, waxy corn, corn, wheat, field pea and lentil starches were autoclaved at 15 psi, 121°C for 1 min. Scanning electron micrographs of the native and autoclaved starches showed no changes in granular surfaces and shapes. In all starches, the X-ray intensities at most of the d-spacings between 3–18 Å increased upon autoclaving, being more pronounced in potato. The X-ray patterns of cereals and legumes remained unchanged, while that of tuber (potato) became more cereal-like. Differential scanning calorimetry of the starch samples revealed that autoclaving increased the gelatinization transition temperatures of wheat but decreased that of potato; the changes observed in waxy corn, corn, field pea and lentil starches were very small. The gelatinization enthalpy of all native starches decreased upon autoclaving while the percentage decrease was highly marked in potato. Image analysis of the native and autoclaved starches revealed changes in the granule size distribution patterns. Also, the population mean area of all native starch granules considerably increased upon autoclaving. Acetyl binding capacity, measured at 5% and 10% acetic anhydride addition levels, was higher in autoclaved than in native starches. Furthermore, autoclaving had no influence on starch cationization, studied at 3% and 6% reagent addition levels. The results indicated that the changes in starch molecular organization caused by autoclaving enhanced its reactivity towards acetylation but not cationization.

**Die Reaktivität von nativen und autoklavierten Stärken von verschiedenen pflanzlichen Herkünften gegenüber Acetylierung und Kationisierung.** Native Kartoffel-, Wachsmais-, Mais-, Weizen-, Felderbsen und Linsenstärken wurden 1 min lang bei 15 psi und 121°C autoklaviert. Die Raster-Elektronenmikrographien der nativen und autoklavierten Stärken zeigten keine Veränderungen der Kernoberflächen und der Gestalt. In allen Stärken waren die Röntgenintensitäten meist der d-Zwischenräume zwischen 3 und 18 Å durch Autoklavierung erhöht und bei Kartoffeln stärker ausgeprägt. Die Röntgenmuster der Cerealien und der Leguminosen blieben unverändert, während die der Knollen (Kartoffel) stärker getreideartig wurden. Die Differential-Raster-Kalorimetrie der Stärkeproben zeigte, daß die Autoklavierung die Verkleisterungs-Temperatur von Weizen erhöht, die von Kartoffeln jedoch erniedrigt wurde; die in Wachsmais-, Mais-, Felderbsen- und Linsenstärken waren dagegen sehr gering. Die Verkleisterungsenthalpie aller nativer Stärken wurde durch Autoklavieren verringert, während sie bei Kartoffeln stark bemerkbar war. Die Bildanalyse der nativen und autoklavierten Stärken ergab Veränderungen im Muster der Korngrößenverteilung. Auch erhöhte sich durch Autoklavieren das Gesamtheitsmittelgebiet aller nativen Stärkekörner beträchtlich. Die Acetylbindungskapazität, gemessen bei 5% und 10% Essigsäureanhydrid-Zusatz war in autoklavierten höher als in nativen Stärken. Weiterhin hatte Autoklavieren keinen Einfluß auf die Stärkekektionisierung, untersucht bei 3% und 6% Reagenzzusätzen. Die Ergebnisse zeigten, daß die durch Autoklavierung verursachten Veränderungen in der molekularen Organisation die Reaktivität gegenüber Acetylierung, jedoch nicht gegenüber Kationisierung steigern.

## 1 Introduction

In order to suit some industrial applications, functional properties of starches are often modified by covalently bonding organic and inorganic chemicals on starch hydroxyl sites. Acetylation, for the textile, paper and food industries, and cationization, for the paper and mining industries, are two important starch chemical modifications [1]. These starch modifications are usually carried out in an aqueous medium and require a high concentration of the modifying reagent to achieve the desired degree of chemical binding. Unreacted reagents are usually removed by repeated washings with water. This unit operation generates substantial volumes of effluent water which are difficult to recycle. Therefore, enhancement of starch granular reactivity prior to chemical modifications may reduce chemical usage as well as the cost of effluent disposal. Heat treatment of starches at low water contents (18–27%) and high temperature (>100°C) in an oven [2–10] or autoclave [11, 12] has been shown to dramatically change the physicochemical properties of starches. In cereal and tuber starches, heat-moisture treatments (by the oven method) increased enzyme susceptibility [7, 10], paste stabilities [8–10] and gelatinization temperatures [5–10], but decreased the swelling power and Brabender peak consistencies [8–10]. Abraham [11] re-

ported similar observations upon autoclaving cassava (root) starches. Holm and Björck [12] showed increased starch availability in wheat flour to  $\alpha$ -amylolysis after dry-autoclaving. It has been postulated that the effects of heat-moisture treatment may be due to molecular reorganization within the crystalline [5] or both crystalline and amorphous [10] regions of starch granules.

The conditions during autoclaving (high temperature, moisture and pressure) are highly conducive for starch gelatinization. However, a short treatment time may not allow the starch granules to equilibrate and reach the minimum moisture content that is necessary for the initiation of gelatinization. The existence of high pressure during autoclaving may intensify the effect of heat on starch granules. The present study was conducted to determine the implications of starch molecular reorganization, caused by autoclaving, on starch granular reactivity towards chemical reagents. The objective was twofold: (a) to investigate the effect of autoclaving on the crystallinity (by X-ray diffractometry), thermal characteristics (by differential scanning calorimetry), surface characteristics and granule size distribution of starches from several plant origins, and (b) to compare the reactivity (degree of chemical binding) of native and autoclaved starch granules towards acetylation and cationization.

## 2 Experimental

### 2.1 Materials

Normal and waxy corn starches were obtained from A. E. Staley Mfg. Co., Decatur, IL, USA. Wheat starch was obtained from Midwest Grain Products, Inc., Atchison, KS, USA. Potato starch was obtained from Sigma Chemical Co., St. Louis, MO, USA. Seeds of lentil (*Lens culinaris* Medic. var. Eston) and field pea (*Pisum sativum* L., var Express) were provided by Dr. A. E. Slinkard, Crop Development Centre, University of Saskatchewan, Saskatoon, SK.

### 2.2 Methods

#### Starch extraction

Extraction of starch from lentil and field pea was carried out according to the procedure of Hoover et al. [13]. All starch samples were thoroughly washed with alkali (starch : 0.02N NaOH = 1:10) and finally with ethyl alcohol (95% w/v) (starch : ethyl alcohol, 1:1.5 w/v), filtered and air-dried overnight.

#### Chemical composition of starch

AACC methods [14] were used to determine moisture (44–19), nitrogen (46–13) and starch damage (76–30A). Starch content was determined by the method of Holm et al. [15] on samples after free sugars were removed by hot ethanol washing (boiled with 80% ethanol for 5 min). Total amylose contents were determined by the method of Chrastil [16]. The lipid contents were determined according to Vasanthan and Hoover [17].

#### Autoclaving

Starch samples (25g, dry weight basis) were weighed into aluminum dishes, evenly spread to a thin bed (2mm thickness) and treated under the following conditions: 15psi, 121°C for 1min (holding time) in an autoclave. Samples were then air dried to a uniform moisture content.

#### X-ray diffraction

X-ray diffractograms were obtained with a Philipps (model 42273) X-ray diffractometer connected to a data acquisition and processing station. Reagent grade alumina powder (Anachemia : 200 mesh  $Al_2O_3$ ) was used as an internal standard (reference peak at 38.50°C 2 $\theta$ ). Traces were obtained using Cu-K $\alpha$  radiation ( $\tau = 1.5478$ ) at 1.6 KVA. The starch powder (0.45 g) was thoroughly mixed with alumina (0.04 g), and packed as densely as possible into an aluminum sample holder. The samples were scanned through the 2 $\theta$  range of 3–40° at an equivalent angular velocity of approximately 0.6° 2 $\theta$  per min. A step interval of 0.01° 2 $\theta$  and a count time of 1 sec was employed. Relative intensities of the starch peaks at different 2 $\theta$  values were obtained from the ratio between the absolute intensity of the starch peak and the reference peak.

#### Differential scanning calorimetry

Starch gelatinization was studied on a Mettler (TA 4000) differential scanning calorimeter equipped with a data acquisition and processing station. Water (21 $\mu$ l) was added with a microsyringe to starch (5.5 mg, db) in aluminum DSC pans which were then hermetically sealed and allowed to stand overnight at room temperature. The scanning temperature range and the heating rate were 20–120°C and 10°Cmin<sup>-1</sup>, respectively. The thermograms were recorded with water as reference. Indium was used for calibration.

#### Scanning electron microscopy

Granule morphology of native and autoclaved starches were studied by scanning electron microscopy. Starch samples were mounted on circular aluminum stubs with double sticky tape, and then coated with gold and examined in a Philipps (SEM 505) scanning electron microscope at an accelerating potential of 20KV.

#### Starch granule size analysis

Mounts for light microscopy of the starch samples were prepared from a dilute slurry of starch/ethanol (95% w/v) mixture. Images of the starch granules from the light microscope (63 x magnification) were analyzed for particle size distribution on a BioQuant System IV image analyzer (R & M Biometrics Inc.), equipped with an image acquisition and processing station. In each measurement, an image of an optical micrometer was introduced for size calibration. Two thousand starch granules, from each native and autoclaved sample, were measured for their diameter (the diameter of a circle of equal area to the particle), and the percentages of particles that occurred within the predetermined diameter ranges were obtained.

#### Acetylation

Acetylation was carried out according to the method of Wurzburg [18]. Acetic anhydride (10 and 5% based on dry weight of starch) was slowly added dropwise while mixing the starch slurry (50g dry wt. starch : 140 ml water), using a magnetic stirrer. Simultaneously 2N NaOH solution containing 15% Na<sub>2</sub>CO<sub>3</sub> was added at a rate sufficient to maintain the pH of the suspension between 8.0–8.4. At completion of the acetic anhydride addition, the pH of the slurry was adjusted to 6.0 with 1N HCl, the starch in the slurry was washed four times with distilled water, recovered by filtration and dried overnight in an oven at 25°C. Acetyl contents of starches were determined according to the Food Chemical Codex [19].

#### Cationization

Cationization was carried out by the aqueous alcoholic alkaline process of Bhirud et al. [20] in a semi-aqueous media. Each sample (10g, dry weight basis) was slurried in 65ml of an aqueous alcoholic alkaline media (50ml of 95% (w/v) ethanol, 15 ml of distilled water and 0.7g of NaOH). Cationic reagent (aqueous solution of 3-chloro-2-hydroxypropyltrimethylammonium chloride (CHTAC) containing 0.57g of active ingredient per ml of reagent) (Aldrich Chemical Company, Inc., Milwaukee, WI, USA) was added (6% and 3% basis of starch dry weight) dropwise while the slurry temperature was maintained at 50°C. The reaction was carried out for 5 h before the slurry was neutralized with 1N HCl. Starch was recovered by filtration, washed five times with 25% (w/c) ethanol, finally washed with 95% (w/v) ethanol and dried overnight in an oven at 30°C. Percentage of cationic groups bound to starch was estimated from the difference in nitrogen content between native and cationized starches.

## 3 Results

#### Chemical composition

The chemical composition of the native starches is presented in Table 1. Starch contents of the samples ranged between 92–96%. Protein (<0.24%), lipid (<0.86%) and ash (<0.26%) contents of all starches were relatively low, indicated that all starches were of high purity. The amylose contents of the starches were: lentil (38.8%) > field pea (31.6%) > wheat (27.4%) > corn (25.6%) > potato (21.2%) > waxy maize (1.2%).

Table 1. Chemical Composition of Native Starches.

Starch source	Composition (% dry starch basis)					
	Moisture	Starch	Protein	Lipid	Ash	Amylose content
Potato	14.1 ± 0.6	95.2 ± 1.8	0.17 ± 0.01	0.13 ± 0.02	0.26 ± 0.04	21.2 ± 0.8
Waxy corn	10.2 ± 0.5	93.0 ± 0.9	0.24 ± 0.05	0.34 ± 0.04	0.11 ± 0.02	1.2 ± 0.4
Corn	9.8 ± 0.6	92.5 ± 1.6	0.21 ± 0.02	0.86 ± 0.11	0.07 ± 0.01	25.6 ± 0.6
Wheat	10.1 ± 1.1	94.2 ± 0.7	0.19 ± 0.01	0.65 ± 0.04	0.20 ± 0.01	27.4 ± 0.4
Field pea	9.5 ± 0.3	96.6 ± 2	0.08 ± 0.02	0.18 ± 0.02	0.12 ± 0.03	31.6 ± 0.6
Lentil	10.0 ± 0.8	95.5 ± 1.1	0.10 ± 0.01	0.23 ± 0.06	0.08 ± 0.01	38.8 ± 0.6

Values are averages of two determinations ± SD.

### Scanning electron microscopy

The granule shapes of the native starches ranged from oval to elliptical to irregular, and were characteristic of each species (Fig. 1). The granule surfaces of all native starches appeared to be smooth. Autoclaving did not cause any significant changes in the granule shapes and surfaces: granular surfaces examined under high magnification (2000x) remained as smooth as those of their native counterparts (not shown).

### X-ray diffraction

The X-ray diffractograms of the native and autoclaved starches are presented in Fig. 2. The crystal interplanar spacings (d-spacings), which are reported [21] to be characteristic of cereal (5.8Å, 5.2Å and 3.8Å; A-type), tuber (16.0Å, 5.9Å, 5.2Å, 4.0Å and 3.7Å; B-type) and legume (16.0Å, 5.8Å, 5.2Å, 3.8Å; C-type) starches, were generally observed in the present starches. However, in field pea and lentil starches, the characteristic peak at 16Å was not observed, which is in agreement with those of Hoover and Sosulski [22, 23] on other legume starches from pinto, navy and black beans. Autoclaving did not cause any significant changes in the X-ray diffractogram patterns or the characteristic d-spacings of native cereal and legume starches. However, in native potato (B-type) starch, changes in the X-ray pattern (Fig. 2 A&B), the disappearance of peak at d-spacing 16.0Å and a shift in other characteristic d-spacings towards to that of cereals (A-type) were observed, which suggested a crystal transformation from B to A type during autoclaving. Similar crystal transformation was reported [9, 10] during heat-moisture treatment of potato starch in an oven.

Graphs showing the differences between relative intensities [relative intensity of autoclaved ( $RI_A$ ) - relative intensity of native ( $RI_N$ )] vs crystal d-spacings between 3–18Å at 0.005Å intervals in X-ray diffraction are presented in Fig. 3. In the six starches, the net relative intensities ( $RI_A - RI_N$ ) at most of the d-spacings were positive, which indicated that autoclaving generally increased the X-ray relative intensities of native starch crystals. In potato, 92% of the total points in the graph were positive. Corresponding values in other starches were: waxy corn, 80%; field pea, 74%; wheat, 72%; lentil, 65%; and corn 55%. The highest and the smallest increases were shown by potato and corn, respectively.

### Particle size analysis

The granule size distributions for the six starch samples varied widely (Fig. 4), ranging from the broad distributions of wheat and potato to the narrow peaks for waxy corn and lentil. Native potato and wheat starches showed trimodal distributions. Autoclaving had a marginal influence on the starch granule size distribution. For potato starch, there was a decrease in proportion of granules in the 8–16 µm size range and an increase at the high range of 17–33 µm. Similar trends were more evident for corn, wheat and field pea starches than for waxy corn and lentil starches. The mean starch granule area and diameter for native and autoclaved starches are given in Table 2.

Table 2. The Mean Area and Diameter of Native and Autoclaved Starch Granules.

Starch source	Treatment	Mean area <sup>a)</sup> (µm <sup>2</sup> )	Mean diameter <sup>a)</sup> (µm)
Potato	Native	653.4 ± 8.9	26.0 ± 0.4
	Autoclaved <sup>b)</sup>	687.0 ± 8.2	27.1 ± 0.4
Waxy corn	Native	78.3 ± 1.4	10.1 ± 0.1
	Autoclaved	94.5 ± 1.8	11.0 ± 0.1
Corn	Native	114.0 ± 2.0	11.4 ± 0.1
	Autoclaved	123.7 ± 2.2	11.8 ± 0.1
Wheat	Native	151.6 ± 4.4	12.5 ± 0.2
	Autoclaved	178.2 ± 4.9	13.9 ± 0.2
Field pea	Native	430.2 ± 6.2	21.2 ± 0.2
	Autoclaved	446.9 ± 5.6	22.7 ± 0.2
Lentil	Native	306.9 ± 4.0	19.3 ± 0.1
	Autoclaved	315.9 ± 3.4	19.7 ± 0.1

<sup>a)</sup> Values are population (2000 granules) mean ± standard error.

<sup>b)</sup> Autoclaved at 121°C, 15 psi for 1 min.

In decreasing order, the mean area (µm<sup>2</sup>) and granule diameter (µm) of each species were: potato, 653.4 and 26; field pea, 430 and 21; lentil, 307 and 19; wheat, 152 and 12.5; corn, 114 and 11.4; and waxy corn, 78.3 and 10.1, respectively. Autoclaving considerably increased the mean area of all native starches (Table 2). The magnitude of increase was highest in potato (33.6 µm<sup>2</sup>) followed by wheat (26.6 µm<sup>2</sup>), field pea (16.7 µm<sup>2</sup>), waxy corn (16.2 µm<sup>2</sup>), corn (9.7 µm<sup>2</sup>) and lentil 9.0 µm<sup>2</sup>.

### Differential scanning calorimetry

The gelatinization transition temperatures (GTT) [ $T_o$  (onset),  $T_p$  (peak) and  $T_c$  (conclusion)] and gelatinization enthalpy ( $\Delta H$ ) of native and autoclaved starches are presented in Table 3. Amongst the six native starches, waxy corn showed the highest GTT (onset, 63.0°C; peak, 73.3°C; and conclusion 83.8°C) followed by corn, field pea, potato, lentil and wheat. Autoclaving increased the GTT of native wheat starch but decreased that of potato starch; the changes observed in corn, waxy corn field pea and lentil starches were negligible. The enthalpies of gelatinization of native starches were: waxy corn (14.5 J/g) > potato (11.3 J/g) > corn (10.2 J/g) > lentil (9.7 J/g) > field pea (7.9 J/g) > wheat (6.8 J/g). Autoclaving decreased the enthalpy of gelatinization ( $\Delta H$ ) in all native starches where the percentage decrease was highest in potato (34.5%) followed by field pea (30.4%), wheat (26.5%), corn (14.7%), lentil (10.3%) and waxy corn (7.5%).

### Acetylation of starches

Addition of 5% acetic anhydride to the reaction mixture with native starches resulted in 1.0 to 1.5% acetylation (Table 4). Doubling of the acetylation reagent increased the degree of acetylation almost proportionately, ranging from 1.8 to 2.8%. Waxy corn was the most reactive starch in the native form, fol-

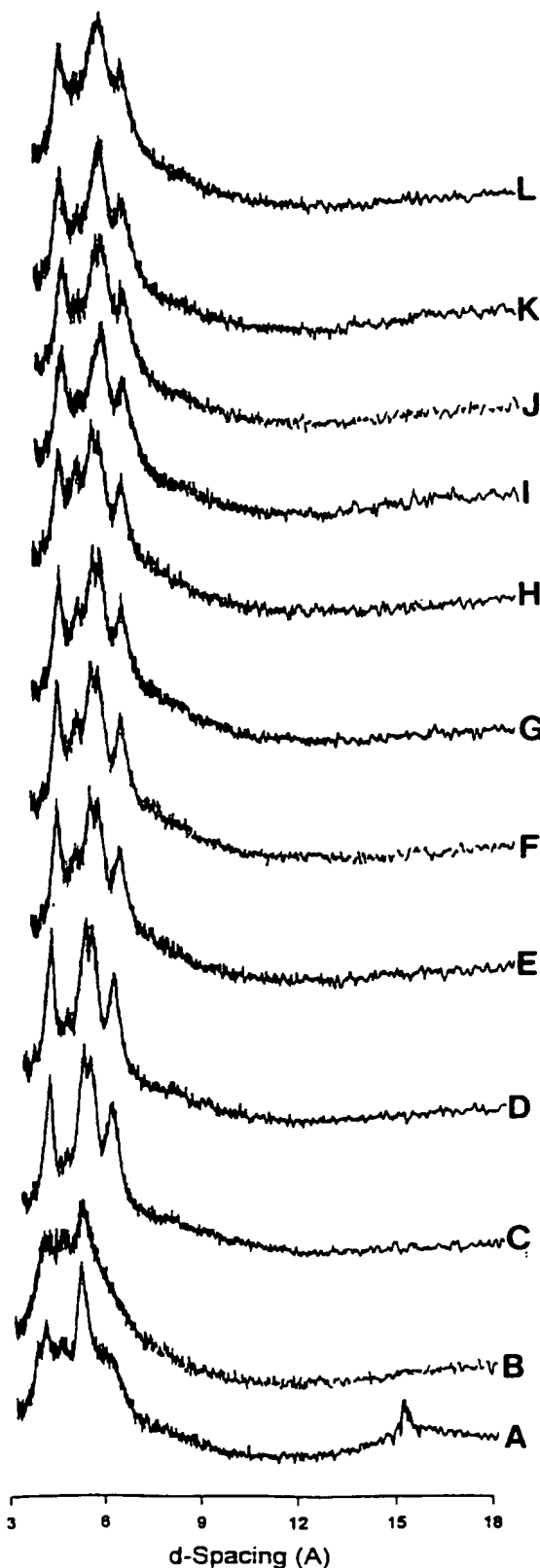


Fig. 2. X-ray diffractograms of native and autoclaved starches. (A) native potato (moisture, 14.1%), (B) autoclaved potato (11.2%), (C) native waxy corn (10.2%), (D) autoclaved waxy corn (10.5%), (E) native corn (9.8%), (F) autoclaved corn (9.5%), (G) native wheat (10.1%), (H) autoclaved wheat (10.5%), (I) native field pea (9.5%), (J) autoclaved field pea (9.6%), (K) native lentil (10.0%) and (L) autoclaved lentil (9.6%).

Table 3. Thermal Characteristics of Native and Autoclaved Starches.

Starch source	Treatment	Transition temperatures (°C) <sup>a</sup>			Enthalpy <sup>b</sup> (J/g)
		Onset	Peak	Conclusion	
Potato	Native	59.1	67.8	77.0	11.3
	Autoclaved <sup>c</sup>	57.8	65.8	74.8	7.4
Waxy corn	Native	63.0	73.3	83.8	14.5
	Autoclaved	62.5	73.3	84.1	13.4
Corn	Native	62.8	71.5	80.5	10.2
	Autoclaved	63.0	71.4	80.7	8.7
Wheat	Native	57.5	64.0	74.0	6.8
	Autoclaved	59.9	66.5	77.6	5.0
Field pea	Native	59.2	69.8	81.8	7.9
	Autoclaved	60.0	70.1	82.2	5.5
Lentil	Native	58.8	65.5	77.2	9.7
	Autoclaved	58.7	65.4	77.8	8.7

Values are mean of three determinations.

<sup>a</sup>) Average SD = 0.5°C

<sup>b</sup>) Average SD = 0.12 J/g.

<sup>c</sup>) Autoclaved at 121°C, 15 psi for 1min.

lowed by potato and other starches. Autoclaving increased the reactivity of the six starch species to acetic anhydride. The percentage increases at 5% and 10% acetic anhydride addition levels were: potato, 68.0% and 61.8%; field pea, 63.8% and 47.2%; wheat, 57.4% and 40.2%; corn, 49.6% and 32.0%; waxy corn, 25.5% and 15%; and lentil, 26.3% and 13.6%, respectively. The descending order observed in percentage acetylation increase was generally similar to that observed in percentage enthalpy decrease due to autoclaving.

#### Cationization of starches

Reaction of 3% and 6% CHTAC with the six native starches resulted in 1.4–1.7% and 1.9–3.2% cationization, respectively (Table 4). However, differences among starch species in degree of cationization were relatively small and autoclaving has no effect on starch reactivity.

## 4 Discussion

Cooke and Gidley [24] have shown by DSC, <sup>13</sup>C-CP-MAS-NMR spectroscopy and powder X-ray diffraction studies that: a) the enthalpy of gelatinization ( $\Delta H$ ) primarily reflects the disruption of double helical order within the starch granules; and b) the integrity of the starch granules is mainly at the double helical rather than the crystalline level. Double helices can exist either in the amorphous or crystalline regions of the native starch granules. The decrease in  $\Delta H$  (Table 3) suggests that autoclaving may have caused a disruption or melting of the double helical structures within the amorphous regions since all starches showed an increase in X-ray relative intensities (Fig. 3) on autoclaving. The disruption of double helices upon autoclaving would expose the starch hydroxyl sites and render them easily accessible for chemical binding.

Hood and Mercier [25] investigated the location of the chemical substituents in modified starch by their effect in blocking action of starch degrading enzymes. These authors concluded that chemical substitution occurred at hydroxyl sites of amylose and more densely around the branching regions (amorphous) of amylopectin molecules. Therefore, it is plausible that the partial disruption of double helices (Table 3) and the consequent development of additional amorphous regions

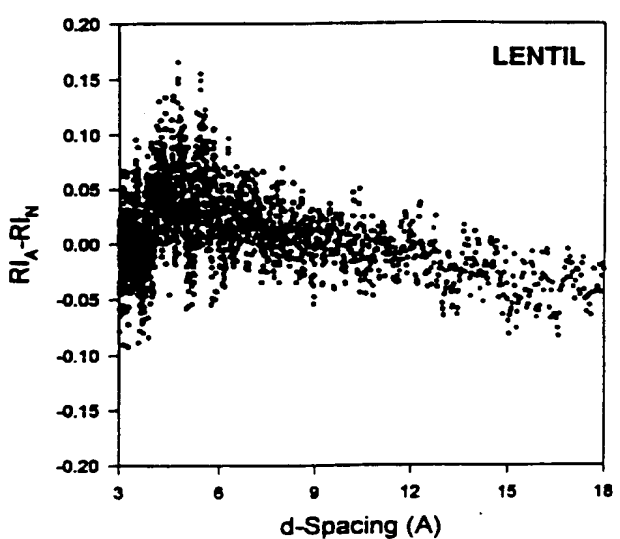
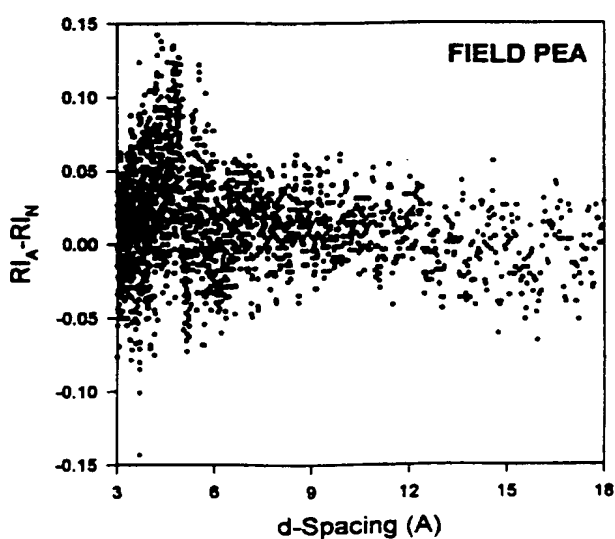
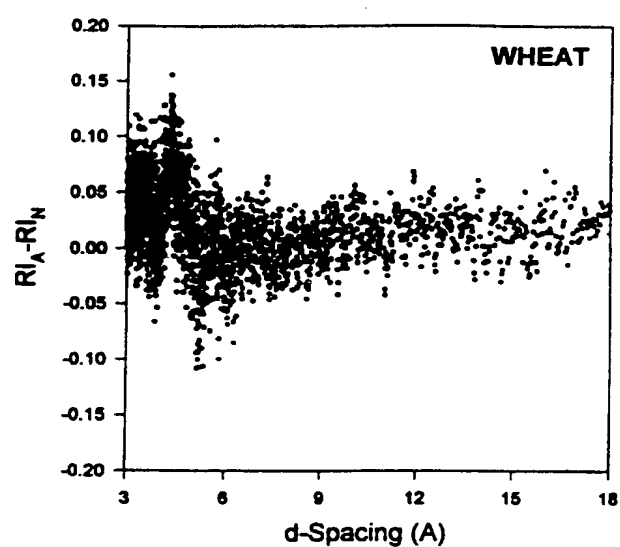
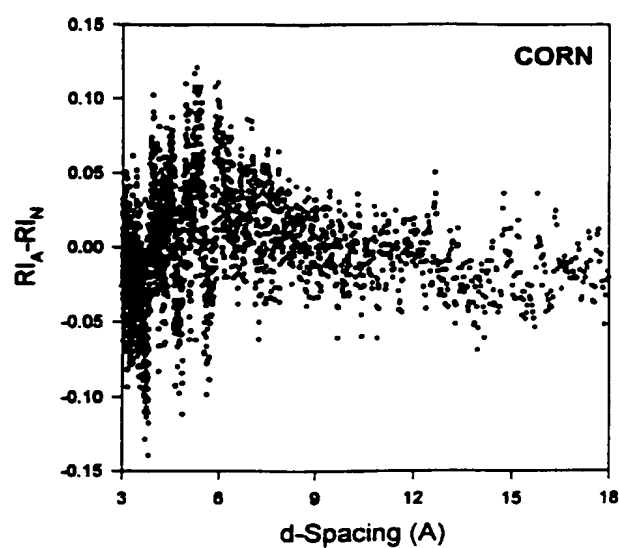
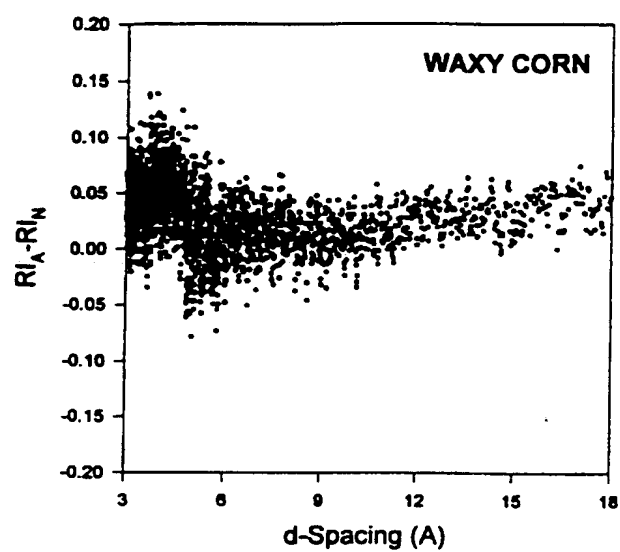
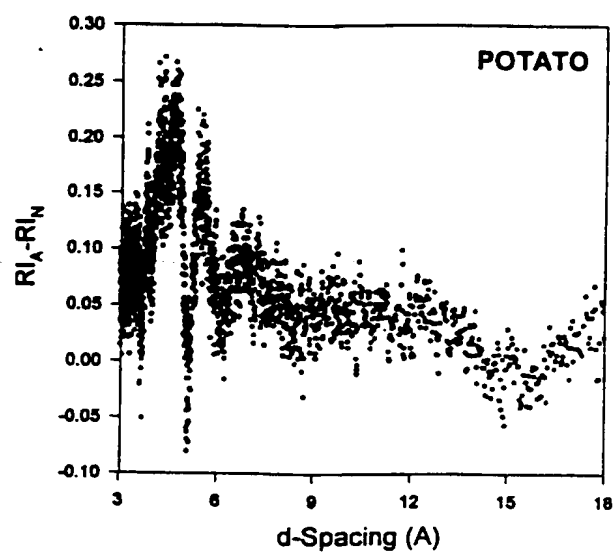


Fig. 3. The differences in the relative intensities between native and autoclaved starches ( $RI_A - RI_N$ ) at d-spacings of 3–18 Å.



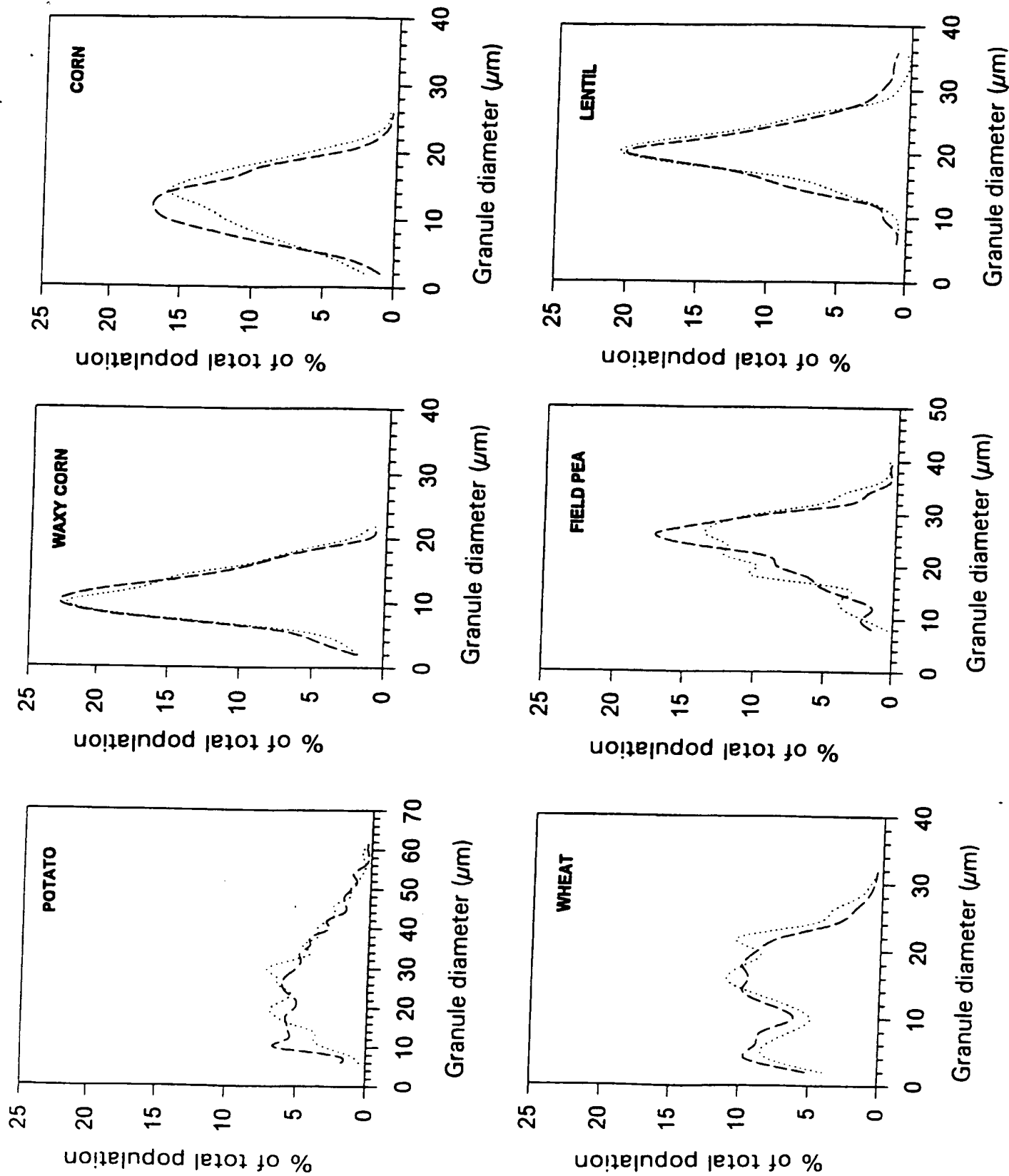


Fig. 4 particle size distribution of native and autoclaved starch granules. - - - Native, . . . . . Autoclaved.

Table 4. Degree of Acetylation and Cationization of Native and Autoclaved Starches.

Starch source	Treatment	Degree of chemical binding (% dry starch basis)			
		Acetic anhydride		CHTAC <sup>a</sup> )	
		(% chemical added)		(% chemical added)	
		5%	10%	3%	6%
Potato	Native	1.22 ± 0.11	2.38 ± 0.07	1.53 ± 0.01	3.19 ± 0.07
	Autoclaved <sup>b</sup> )	2.05 ± 0.07	3.85 ± 0.13	1.47 ± 0.02	2.95 ± 0.02
Waxy corn	Native	1.49 ± 0.12	2.80 ± 0.13	1.60 ± 0.02	2.89 ± 0.05
	Autoclaved	1.87 ± 0.03	3.22 ± 0.15	1.54 ± 0.01	2.80 ± 0.02
Corn	Native	1.13 ± 0.11	2.28 ± 0.17	1.68 ± 0.05	2.82 ± 0.28
	Autoclaved	1.69 ± 0.15	3.01 ± 0.04	1.72 ± 0.02	2.76 ± 0.12
Wheat	Native	1.01 ± 0.14	1.99 ± 0.12	1.42 ± 0.01	2.75 ± 0.04
	Autoclaved	1.59 ± 0.12	2.79 ± 0.06	1.39 ± 0.04	2.70 ± 0.06
Field pea	Native	1.08 ± 0.01	1.78 ± 0.01	1.37 ± 0.02	1.91 ± 0.21
	Autoclaved	1.77 ± 0.18	2.62 ± 0.12	1.44 ± 0.05	1.67 ± 0.03
Lentil	Native	1.29 ± 0.17	2.19 ± 0.06	1.65 ± 0.01	2.59 ± 0.26
	Autoclaved	1.63 ± 0.07	2.49 ± 0.13	1.58 ± 0.02	2.23 ± 0.15

Values are means of three determinations ± SD.

<sup>a</sup>) 3-Chloro-2-hydroxypropyltrimethylammonium chloride.

<sup>b</sup>) Autoclaved at 121°C, 15 psi for 1 min.

around the branching points of amylopectins may have increased the chemical binding ability (reactivity) of autoclaved starches.

The results from the X-ray diffraction study showed that autoclaving increased the relative intensities ( $RI_A - RI_N > 0$ ) at most of the d-spacings (Fig. 3). The increased intensities could mean either the formation of new crystalline regions from the existing double helices in the amorphous regions of the native granules or the perfection of starch crystals that were already existing in the native granules. The formation of new starch crystals seems to be unlikely since the results from DSC indicated that the double helices were melting during autoclaving (double helices are the building blocks of starch crystals [26–28]). Therefore, crystal perfection is more likely to be the reason for the X-ray intensity increase. This means that autoclaving leads to a better organization of crystals that are already existing in the native starch granules; better crystal organization would provide less hindrance for easy passage of chemicals into the granules and may result in a better chemical binding.

Particle size analysis of native and autoclaved starch granules (Fig. 4) suggested that autoclaving transforms some starch granules of smaller size (lower diameter) into larger size (higher diameter). This is probably due to impaired starch granular integrity, caused by the partial disruption of starch double helices (Table 3), which may have facilitated a certain degree of irreversible swelling of starch granules in the presence of heat and moisture during autoclaving. Swelling of starch granules would result in an increase in the granular surface area (Table 2); this would then increase the chemical contact area and thereby enhance chemical binding to starch hydroxyl sites. Furthermore, increased starch granular surface area may improve surface porosity which would facilitate easy penetration of chemicals into the granules.

These results suggest that the increased acetyl group binding of autoclaved starches (Table 4) was mainly due to three factors: a) disruption of double helices which exposes additional hydroxyl sites for chemical binding; b) better crystal organization which may have reduced the hindrance for easy chemical passage into the granules and c) increased granular surface area per unit weight of starch. Marginal differences observed in the percentage of CHTAC binding between native and auto-

claved starches, at both 3% and 6% levels (Table 4) are probably due to very high alkaline (pH > 12) condition that existed in the reaction mixture during cationization. High alkaline conditions would lead to high degrees of starch granular swelling (alkali swelling) which may over-ride the effects of autoclaving. Since acetylation of starches is carried out at mild alkaline conditions (pH 8.0–8.5), the degree of starch granule swelling caused by alkali became negligible and thus the effects of autoclaving were pronounced.

This work has demonstrated that autoclaving of starches would enhance the reaction efficiency of acetylation but not cationization. This means that, in order to achieve a certain degree of acetyl binding (degree of substitution), autoclaved starches will require lower acetic anhydride concentrations in the reaction mixture than their native counterparts. Consequently, at the end of the reaction, the amount of unreacted chemical (residual chemical) in the reaction mixture would be lower and could be removed with less washing, resulting in less effluent water. However, the foregoing beneficial aspects of autoclaving should be evaluated against the cost of autoclaving.

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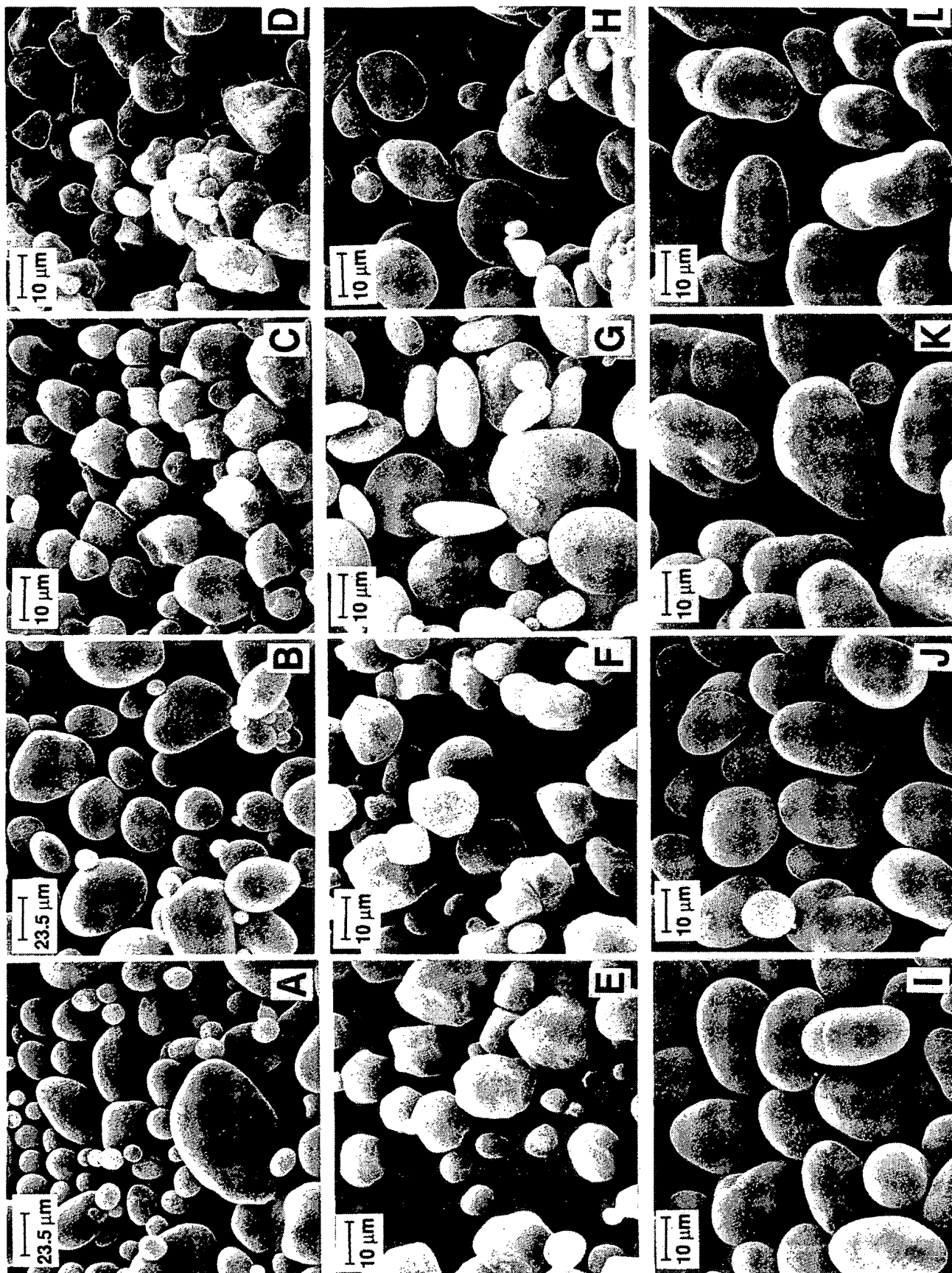


Fig. 1. Scanning electron micrographs of native and autoclaved starches. (A) native potato, (B) autoclaved potato, (C) native waxy corn, (D) autoclaved waxy corn, (E) native corn, (F) autoclaved corn, (G) native wheat, (H) autoclaved wheat, (I) native field pea, (J) autoclaved field pea, (K) native lentil (L) autoclaved lentil.

## LES PROCEDES D'EXTRACTION

### L'USIPA

Un peu d'histoire  
Le conseil de direction  
Les adhérents  
Produits et marchés

### L'AMIDONNERIE FECULERIE

L'amidon : un ami naturel  
> Les procédés d'extraction  
Un secteur dynamique  
Une agro-industrie de pointe

### LES PRODUITS AMYLACES

Les amidons  
Les amidons modifiés  
Le glucose  
Les coproduits  
Les valorisations non-alimentaires

Le métier de l'amidonnerie-féculerie consiste à séparer les constituants de la plante: l'amidon, la protéine, l'enveloppe cellulosique, les fractions solubles et dans le cas du maïs, le germe dont sera extraite l'huile. Cette première phase fait intervenir une série d'étapes simples de séparation physique des constituants: broyage, tamisage, centrifugation etc... A noter toutefois que les procédés de fabrication sont spécifiques à chaque plante et que les outils industriels sont dédiés à une matière première (blé, maïs ou pomme de terre en Europe) sans qu'il y ait donc possibilité de substituer l'une à l'autre. Dans un deuxième temps, l'amidon ainsi extrait sous sa forme la plus pure sera ou bien valorisé "tel quel" après séchage (on parlera "d'amidon natif") ou bien envoyé dans de multiples "ateliers" où il subira diverses transformations visant soit à en améliorer les performances ("amidons modifiés") soit à obtenir les sirops de glucose. Ces derniers donneront à leur tour naissance à de multiples produits finis... mais c'est une autre histoire!

Au final l'amidon et les produits dérivés seront livrés aux industries clientes sous forme de poudre (à l'image de la farine) ou de sirops.

A noter qu'en français, contrairement à l'anglais qui n'utilise qu'un seul terme (starch), l'amidon est appelé "féculé" quand il est extrait des tubercules ou des racines (pommes de terre, manioc ou patates douces).



Analyse des

# STARCH:

## Chemistry and Technology

EDITED BY

**ROY L. WHISTLER**

DEPARTMENT OF BIOCHEMISTRY  
PURDUE UNIVERSITY  
LAFAYETTE, INDIANA

**EUGENE F. PASCHALL**

MOFFETT TECHNICAL CENTER  
CORN PRODUCTS CO.  
ARGO, ILLINOIS

ASSISTANT EDITORS

**J. N. BEMILLER**

DEPARTMENT OF CHEMISTRY  
SOUTHERN ILLINOIS UNIVERSITY  
CARBONDALE, ILLINOIS

**HUGH J. ROBERTS**

KRAUSE MILLING COMPANY  
MILWAUKEE, WISCONSIN

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larger:  $15.0\ \mu$  on a number basis and  $17.4\ \mu$  on a weight basis compared with respective values of  $9.2\ \mu$  and  $14.1\ \mu$  for corn starch (112). A more significant difference between these two starches is that milo starch has a gelatinization temperature range of  $67^{\circ}$ – $75^{\circ}$ , which is about  $3^{\circ}$  higher than the  $62^{\circ}$ – $72^{\circ}$  range observed with corn starch (112). Waxy milo

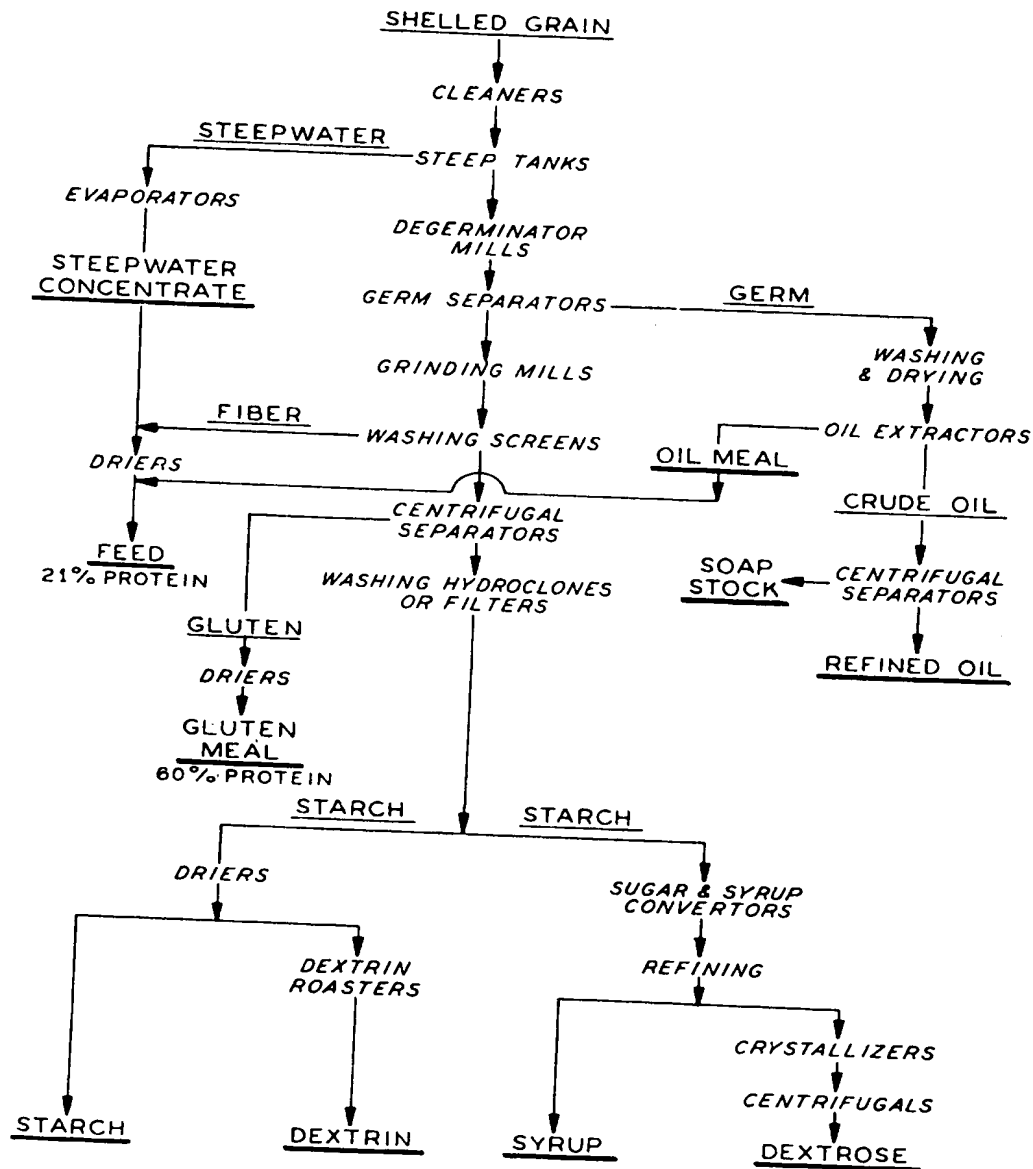


FIG. 8.—Flow diagram of the wet milling process.

starch gelatinizes at a temperature about  $5^{\circ}$  higher than nonwaxy milo starch, a characteristic of all waxy starches relative to their nonwaxy counterparts. Paste properties of milo starches are similar to those of corn starches (111, 113).

#### IV. THE WET MILLING PROCESS

Wet milling of corn and milo for starch manufacture is more than simple grinding of water soaked grain. It is a highly specific and completely



The fiber that cannot pass through the slots is discharged through a separate outlet. Several centrifugal sieves are usually employed in series. An advantage over ordinary screen separation is that the solids' content of the waste pulp from the centrifugal sieve is about three times that of pulp from the conventional screen. In the operations shown in Figure 2, the extracted pulp having 13% solids' content is pressed to 23% solids and then dried to provide a component for livestock feed.

#### Purification and separation

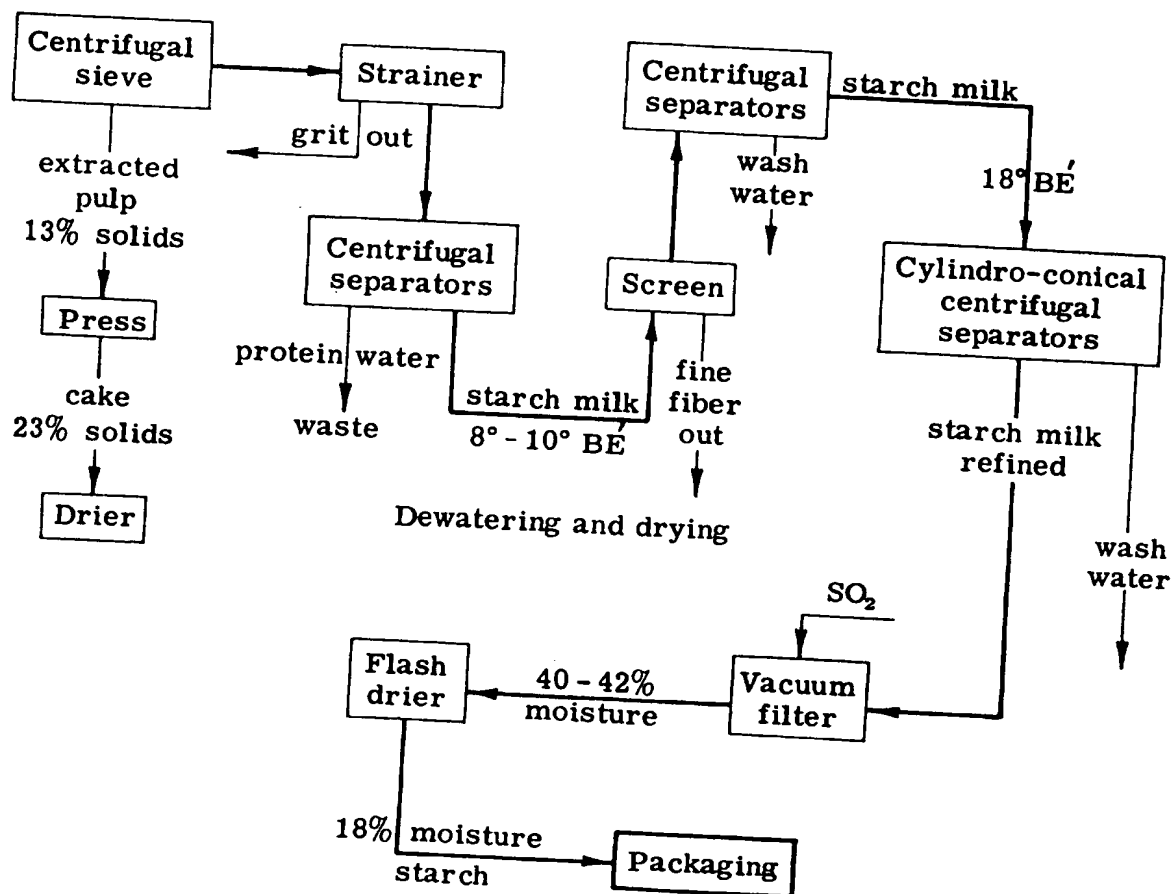


FIG. 2.—Detail of steps in separation, purification, dewatering, and drying potato starch. From Talburt and Smith (6).

Centrifugal separators operating at high speed are used to wash the starch milk received from the rotary sieves. Figure 4 presents a cut-away diagram of a centrifugal separator. In centrifugal refining, the starch milk passes through the separator bowl in which starch grains are removed from the remaining suspension by continuous ejection through nozzles in the bowl wall. The protein water effluent is replaced to a considerable extent by clean water. Centrifugal separators can also act as concentrators. Thus, in the Figure 2 example, the starch milk leaving the first centrifugal separator at 8°-10° Bé is increased to 18° Bé by passage through the second separator.